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ORIGINAL RESEARCH

Long-Term Hepatic and Extrahepatic Outcomes of Chronic Hepatitis C Patients After Sofosbuvir-Based Treatment (LONGHEAD Study)

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ABSTRACT

***Background/Aims*:** Direct-acting antivirals (DAAs) are highly effective in treating hepatitis C virus (HCV) infection. The long-term hepatic and extrahepatic outcomes of DAAs in chronic hepatitis C (CHC) patients receiving curative antivirals are elusive.

***Methods*:** CHC patients were retrieved from two phase III sofosbuvir-based clinical trials conducted from 2013–2014. Patients who achieved a sustained virological response have been followed prospectively for 5 years since 2016. A propensity score-matched interferon-based historical control with a 1:3 ratio was used for comparison. Quality of life (QoL) was measured by the SF-36, liver fibrosis was measured by

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electrography, and fibrosis-related markers were followed annually in the prospective cohort.
Results: A total of 160 DAA- and 480 interferon-treated patients were enrolled. Twenty-eight patients developed hepatocellular carcinoma (HCC) over a follow-up period of 4424 person-years (annual incidence: 0.6%). The incidence of HCC did not differ significantly between the DAA cohort and interferon-treated patients ($P=0.07$). Cox regression analysis revealed that FIB-4 was the only factor independently associated with HCC development (hazard ratio [HR]: 95% confidence interval [CI] 3.59/1.68–7.66, $P=0.001$). The incidence of newly developed cardio-cerebrovascular disease was 13.8 per 1000 person-years and 0.9 per 1000 person-years in interferon-treated patients and the DAA cohort, respectively ($P<0.001$). Interferon-based patients had a significantly greater incidence of cardio-cerebrovascular disease (HR/CI 3.39/1.28–8.96, $P=0.014$). There was a substantial decrease in liver stiffness ($P_{trend}=0.08$) and M2BPGi ($P_{trend}=0.05$) and a significant reduction in LOXL2 ($P_{trend}=0.02$) over 5 years. A significant decrease in QoL was observed in role limitations due to physical health and emotional problems, whereas the other parameters were maintained consistently throughout the 5 years of follow-up.
Conclusions: HCV eradication by DAAs improved liver- and non-liver-related outcomes, constantly promoted liver fibrosis regression, and maintained quality of life after HCV cure.
Clinical Trial Number: NCT03042520.

Keywords: DAA; HCV; SVR; Long-term outcome

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Key Summary Points

Why carry out this study?

Direct-acting antivirals (DAAs) are highly effective in treating hepatitis C virus (HCV) infection.

The long-term hepatic and extrahepatic outcomes of DAAs in chronic hepatitis C (CHC) patients receiving DAAs are lacking using a prospective design.

What was the hypothesis of the study?

The long-term liver and non-liver related outcomes are favorable in CHC patients with curative DAAs.

What was learned from the study?

HCV eradication by DAAs improves liver-related outcomes, decreased diabetes and cardiocerebral vascular incidences.

HCV eradication constantly promotes liver fibrosis regression.

CHC patients with curative DAAs maintain their long-term quality of life up to 5 years.

INTRODUCTION

An estimated 50 million people are infected by hepatitis C virus (HCV) globally, and chronic hepatitis C (CHC)-related liver and non-liver consequences remain a major disease burden for humans. The World Hepatitis Organization has set up interim guidance for HCV control [1]. With the ambitious goal of HCV elimination by 2030, most countries are forecasted as not on track to achieve global elimination targets [2].

Successful HCV eradication significantly promotes liver fibrosis regression and reduces the risk of liver-related events and extrahepatic outcomes [3–9]. The development of highly effective direct-acting antiviral agents (DAAs) has greatly facilitated the treatment of HCV infection. Compared with interferon-based regimens, DAAs have an equal benefit in reducing the risk of hepatocellular carcinoma (HCC) [10, 11]. Notably, reports of the long-term benefits of sustained virological response (SVR) were largely retrieved from retrospective cohorts, in which selection bias may exist and heterogeneous patient characteristics from real-world data may confound the outcome of measurements. To overcome this pitfall, in this study, we strictly followed a DAA-treated cohort prospectively from two phase III clinical trials for 5 years. We aimed to address liver-related outcomes in terms of HCC development, liver-related events and potential fibrosis regression, as well as extrahepatic outcomes, including the occurrence of diabetes, cardio-cerebrovascular disease, and long-term changes in quality of life.

METHODS

Patients

CHC patients were retrieved from two phase IIIb clinical trials (ledipasvir/sofosbuvir for HCV genotype 1 (HCV-1) [GS-US-337-0131] and sofosbuvir + ribavirin for HCV genotype 2 [GS-US-334-0115]) [12–15], which were conducted at 24 sites in Taiwan [12, 13] and Korea [14, 15] from December 2013 to November 2014. The participants were prospectively followed semiannually from October 2016 until November 2023. Patients were excluded if they had a history of liver decompensation, hepatocellular carcinoma (HCC), or hepatitis B virus dual infection, if they failed to achieve an SVR, or if the outcome of interest occurred before treatment or within 1 year after the end of antiviral therapy. An interferon-based historical controlled cohort, T-COACH (Study on Taiwanese Chronic Hepatitis C Cohort; from January 2003 to December 2014) [4], was included to compare liver- and non-liver-related outcomes. They were propensity score-matched for age, sex, liver fibrosis, and follow-up period at a 1:3 ratio. The clinical events were recorded with a designated case report form in the DAA cohort and were reviewed from the medical chart in the interferon-based control group. The study was approved by the institutional review board of Kaohsiung Medical University Hospital

(KMUHIRB-F(I)-20160082), which conforms to the guidelines of the International Conference on Harmonization for Good Clinical Practice and was performed in accordance with the Helsinki Declaration of 1964 and its later amendments. All patients provided written informed consent.

The primary objective was to compare the incidence of HCC and liver-related outcomes (LROs) between interferon-treated and DAA-treated cohorts. The secondary objective was to compare the extrahepatic outcomes, including newly developed diabetes and cardio-cerebrovascular disease, between groups as well as the long-term changes in liver fibrosis and quality of life (QoL) in the DAA-treated cohort. LROs included hepatic encephalopathy, esophageal/gastric variceal bleeding or newly developed ascites, jaundice, or liver dysfunction-related coagulopathy. Cardio-cerebrovascular diseases included ischemic or hemorrhagic stroke, angina pectoris, and myocardial infarction.

Laboratory Analysis

Baseline patient characteristics in the DAA-treated cohort were retrieved from parent studies [12–15]. Liver fibrosis was prospectively measured by transient elastography (FibroScan®; Echosens, Paris, France), serum Mac-2 binding protein glycosylation isomer (M2BPGi), and lysyl oxidase-like-2 (LOXL2) annually for up to 5 years. M2BPGi was quantified via a lectin-antibody sandwich immunoassay with a fully automatic HISCL-5000 immune analyzer (Sysmex Co., Hyogo, Japan) [16]. LOXL2 levels were measured using an enzyme-linked immunosorbent assay (ELISA) kit (Cloud-Clone Corp., TX). SVR was defined as undetectable HCV RNA (< 12 IU/ml or < 25 IU/ml depending on individual laboratory testing) throughout 12 weeks post-DAA and 24 weeks post-interferon-based treatment. Liver cirrhosis was defined as transient elastography > 12 kPa [17] or clinical evidence of cirrhotic portal hypertension. Newly developed HCC was confirmed by pathology or clinical judgment based on the regional guidelines [18]. LRO included HCC, liver decompensation, variceal bleeding, and hepatic encephalopathy. Quality of life (QoL) was evaluated annually using the Short Form Health Survey (SF-36) questionnaire [19].

Statistical Analyses

Frequencies were compared between groups using the χ^2 test with Yates' correction or Fisher's exact test. The group means (presented as the mean ± standard deviations) were compared using analysis of variance and Student's *t*-test or the nonparametric Mann-Whitney test when appropriate. The fibrosis index 4 (FIB-4) was calculated using the following formula: age (years) × aspartate aminotransferase (AST in U/l)/(platelets in 10^9/l) × (alanine transaminase [ALT in U/l])$^{1/2}$. A Cochran-Armitage test was performed to compare the trends in the changes in QoL and liver fibrosis-related markers over time. A Kaplan-Meier analysis and log-rank test were performed to compare the cumulative incidence of liver and non-liver-related outcomes between groups. Cox regression analysis was applied to analyze the factors independently associated with the outcome of interest by analyzing the covariates with *P* values < 0.1 in the univariate analysis. The statistical analyses were performed using the SPSS 12.0 statistical package (SPSS, Chicago, IL, USA). All the statistical analyses were based on two-sided hypothesis tests with a significance level of $P < 0.05$.

RESULTS

Patients

A total of 200 DAA-treated patients from the parent studies were initially included. Eighty-eight (44%) HCV-1 patients received ledipasvir/sofosbuvir, and 112 (56%) HCV-2 patients received sofosbuvir + ribavirin. One hundred sixty DAA-treated patients with long-term outcomes available and 480 interferon-based controls were included for comparison (Supplementary Fig. 1). In the DAA cohort, the mean age was 55.7 years, and males accounted for

Table 1 Patient characteristics before antiviral therapy

Variables	Interferon-based cohort ($n=480$)	DAA cohort ($n=160$)	*P* value
Age, years	55.7 ± 9.0	55.7 ± 8.4	0.97
Male gender	191 (39.8)	61 (38.1)	0.71
BMI, kg/m^2	25.0 ± 3.6	24.5 ± 3.2	0.12
Diabetes	87/479 (18.2)	22/160 (13.8)	0.20
Hypertension	141/480 (29.4)	40/160 (25.0)	0.29
Dyslipidemia	76/480 (15.8)	22/160 (13.8)	0.53
HCV genotype 1	183 (38.9)	68 (42.5)	0.43
HCV RNA, log^{10} IU/ml	5.5 ± 1.0	6.4 ± 0.8	< 0.01
≥ 2,000,000 IU/ml	118 (24.6)	101 (63.1)	< 0.01
AST ratio, ULN	1.9 ± 1.3	1.4 ± 1.0	< 0.01
ALT ratio, ULN	2.8 ± 2.3	2.0 ± 1.8	< 0.01
Platelet count, × 10^3/μl	185.8 ± 50.9	187.3 ± 63.5	0.79
FIB-4	2.5 ± 1.8	2.5 ± 2.1	0.86
> 3.25	78 (16.3)	30 (18.8)	0.46
Creatinine, mg/dl	0.9 ± 0.9	0.8 ± 0.2	< 0.01
eGFR, ml/min/1.73 m^2	96.6 ± 29.8	100.1 ± 19.0	0.09
< 60	35 (7.3)	2 (1.3)	< 0.01
Fasting sugar, mg/dl	103.5 ± 33.3	104.7 ± 35.9	0.74
Follow-up duration, years	7.4 ± 3.6	7.3 ± 1.4	0.52

Variables are expressed as mean ± standard deviation or sample size and proportion (%)

DAA directly acting antivirals, *BMI* body mass index, *AST* aspartate aminotransferase, *ALT* alanine aminotransferase, *r-GT* r-glutamyl transferase, *ULN* upper limit normal, *FIB-4* fibrosis-4 index, *eGFR* estimated glomerular filtration rate

38.1% ($n=61$) of the population. The mean FIB-4 score was 2.5 ± 2.1 before antiviral therapy. Compared with the DAA cohort, a smaller proportion of interferon-based patients had HCV-2, a lower HCV RNA level, and higher levels of liver enzymes and serum creatinine (Table 1).

Liver-Related Outcomes

Twenty-eight patients developed HCC over a follow-up period of 4424 person-years (annual incidence: 0.6%). The 1-, 3-, and 5-year cumulative incidence rates of HCC were 0%, 1.2%, and 2.3%, respectively, in interferon-treated patients compared with 0%, 0%, and 0.7%, respectively, in the DAA cohort (log-rank $P=0.07$). Compared with patients without HCC development, patients with HCC were older and had lower platelet counts and higher AST and FIB-4 levels. Cox regression analysis revealed that FIB-4 was the only factor independently associated with HCC development (hazard ratio [HR]: 95% confidence interval [CI] 3.59/1.68–7.66, $P=0.001$) (Table 2).

Table 2 Factors associated with HCC development

Variables	HCC (+) ($n=28$, 4.6%)	HCC (−) ($n=581$, 95.4%)	*P* value	Cox-regression analysis	
				HR (95% CI)	*P* value
Age, years	58.6 ± 7.4	55.3 ± 8.9	0.03		
Male gender	15 (53.6)	219 (37.7)	0.09		
BMI, kg/m^2	25.7 ± 4.2	24.8 ± 3.5	0.30		
Diabetes	7 (25.0)	92 (15.9)	0.20		
Hypertension	12 (42.9)	151 (26.0)	0.05		
Dyslipidemia	7 (25.0)	86 (14.8)	0.14		
HCV genotype 1	11 (42.3)	226 (39.9)	0.76		
HCV RNA, ≥ 2,000,000 IU/ml	6 (21.4)	206 (35.5)	0.13		
AST ratio, > ULN	2.3 ± 1.2	1.7 ± 1.2	0.03		
ALT ratio, > ULN	2.7 ± 1.5	2.6 ± 2.2	0.59		
r-GT ratio, > ULN	1.2 ± 1.2	0.7 ± 0.7	0.11		
Platelet count, × 10^3/μl	162.4 ± 50.4	188.6 ± 54.4	0.01		
FIB-4, > 3.25	11 (39.3)	86 (14.8)	< 0.01	3.59 (1.68–7.66)	0.001
Creatinine, mg/dl	0.9 ± 0.4	0.9 ± 0.8	0.65		
DAA/interferon-based regimen	3 (10.7)/25 (89.3)	156 (26.9)/425 (73.2)	0.08		

Variables are expressed as mean ± standard deviation or sample size and proportion (%)

Age, AST, ALT, and platelets were not put into multivariate analysis because of strong collinearity with FIB-4

HCC hepatocellular carcinoma, *HR* hazard ratio, *CI* confidence interval, *BMI* body mass index, *AST* aspartate aminotransferase, *ALT* alanine aminotransferase, *r-GT* r-glutamyl transferase, *ULN* upper limit normal, *FIB-4* fibrosis-4 index, *DAA* directly acting antivirals

In addition to those with HCC, four patients (2 with liver decompensation, 1 with esophageal variceal bleeding, and 1 with hepatic encephalopathy) in the interferon-treated group but none in the DAA cohort experienced LROs after achieving an SVR. In total, the 1-, 3-, and 5-year cumulative incidence rates of LROs were 0%, 1.4%, and 2.9%, respectively, in interferon-treated patients compared with 0%, 0%, and 0.7%, respectively, in the DAA cohort (log-rank $P=0.04$). Patients with LROs were older and were more likely to be male, have hypertension, receive interferon-based therapy, have lower platelet counts, and have higher AST and FIB-4 levels. Cox regression analysis revealed that the factor most strongly associated with LRO development was FIB-4 (HR: 4.77, CI 2.36–9.64, $P<0.001$), followed by male sex (HR/CI 3.65/1.76–7.55, $P=0.001$) and hypertension (HR: 2.40, CI 1.17–4.90, $P=0.02$) (Table 3). Neither the HCC nor the LRO differed significantly between the DAA cohort and the interferon-treated patients. The pre-DAA FIB-4 scores of the 3 HCC patients were 1.1, 3.6, and 4.3, respectively. The evolution of liver fibrosis among the three HCC patients and their counterparts in the DAA cohort is shown in Supplementary Fig. 2. There was an increasing trend in liver stiffness in HCC patients, whereas there was no significant difference in the values of noninvasive seromarkers between the groups.

Table 3 Factors associated with liver-related outcomes (LRO)

Variables	LRO (+) (*n* = 32, 5.3%)	LRO (−) (*n* = 574, 94.7%)	*P* value	Cox regression analysis	
				HR (95% CI)	*P* value
Age, years	59.1 ± 7.2	55.2 ± 8.9	0.01		
Male gender	19 (59.4)	214 (37.3)	0.01	3.65 (1.76–7.55)	0.001
BMI, kg/m^2	25.7 ± 4.0	24.8 ± 3.4	0.25		
Diabetes	9 (28.1)	89 (15.5)	0.06		
Hypertension	14 (43.8)	149 (26.0)	0.03	2.40 (1.17–4.90)	0.02
Dyslipidemia	8 (25.0)	84 (14.6)	0.11		
HCV genotype 1	17 (56.7)	346 (60.9)	0.64		
HCV RNA, ≥ 2,000,000 IU/ml	7 (21.9)	205 (35.7)	0.11		
AST ratio, > ULN	2.5 ± 1.5	1.7 ± 1.2	0.01		
ALT ratio, > ULN	2.9 ± 1.6	2.6 ± 2.2	0.33		
r-GT ratio, > ULN	1.2 ± 1.2	0.7 ± 0.7	0.05		
Platelet count, × 10^3/μl	156.3 ± 51.7	189.0 ± 54.2	< 0.01		
FIB-4, > 3.25	14 (43.8)	83 (14.5)	< 0.01	4.77 (2.36–9.64)	< 0.001
Creatinine, mg/dl	0.9 ± 0.4	0.9 ± 0.8	0.72		
DAA/ interferon-based regimen	3 (9.4)/29 (90.6)	156 (27.2)/418 (72.8)	0.03	3.04 (0.88–10.54)	0.08

Variables are expressed as mean ± standard deviation or sample size and proportion (%)

Age, AST, ALT, and platelets were not put into multivariate analysis because of strong collinearity with FIB-4

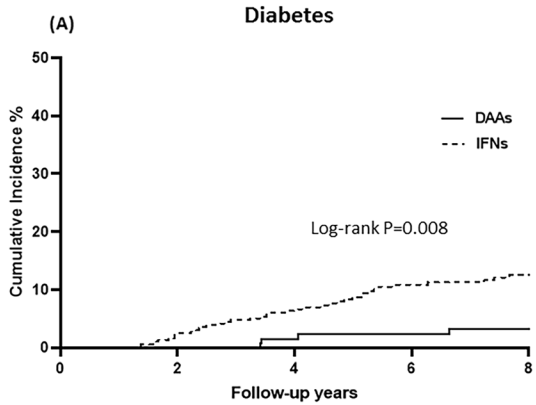
LRO liver-related outcome, *HR* hazard ratio, *CI* confidence interval, *BMI* body mass index, *AST* aspartate aminotransferase, *ALT* alanine aminotransferase, *r-GT* r-glutamyl transferase, *ULN* upper limit normal, *FIB-4* fibrosis-4 index, *ULN* upper limit normal, *DAA* directly acting antivirals

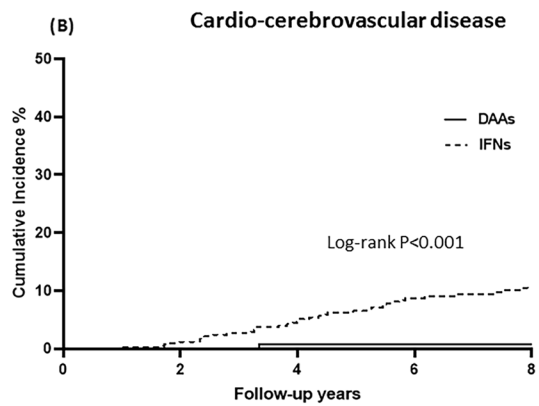
Non-liver-Related Outcomes

Characteristics of patients without underlying diabetes and cardio-cerebrovascular disease before antiviral therapy are shown in Supplementary Table 1 and Supplementary Table 2. The incidence of newly developed diabetes was 17.2 per 1000 person-years and 4.8 per 1000 person-years in interferon-treated patients and the DAA cohort, respectively (log-rank $P < 0.001$). Cox regression analysis revealed that interferon-based patients had a significantly greater incidence of diabetes (HR/CI 3.39/1.28–8.96, $P = 0.014$) (Fig. 1A). The incidence of newly developed cardio-cerebrovascular disease was 13.8 per 1000 person-years and 0.9 per 1000 person-years in interferon-treated patients and the DAA cohort, respectively (log-rank $P < 0.001$). Cox regression analysis revealed that interferon-based patients had a significantly greater incidence of cardio-cerebrovascular disease (HR/CI 12.6/1.70–93.18, $P = 0.013$) (Fig. 1B).

Long-Term Changes in Liver Fibrosis and QoL in the DAA Cohort

As shown in Table 4, the FIB-4 score did not change drastically during the follow-up period. In contrast, there was a substantial decrease in liver stiffness ($P_{trend} = 0.08$) and M2BPGi levels ($P_{trend} = 0.05$) and a significant reduction in LOXL2 levels ($P_{trend} = 0.02$) over 5 years.





Group		BL	Cumulative Incidence (%)				Crude HR (95% CI) P value	*Adjusted HR (95% CI) P value
			1Y	3Y	5Y	7Y		
IFN	No. at risk (%)	393	386 (0.0)	323 (4.7)	263 (8.7)	215 (11.2)	3.28 (1.29-8.34) P=0.013	3.39 (1.28-8.96) P=0.014
DAA	No. at risk (%)	138	138 (0.0)	138 (0.0)	120 (2.3)	106 (3.2)	1	1

Group		BL	Cumulative Incidence (%)				Crude HR (95% CI) P value	**Adjusted HR (95% CI) P value
			1Y	3Y	5Y	7Y		
IFN	No. at risk (%)	464	454 (0.0)	381 (2.6)	314 (6.5)	264 (9.4)	15.64 (2.15-114.0) P=0.007	12.60 (1.70-93.18) P=0.013
DAA	No. at risk (%)	157	157 (0.0)	157 (0.0)	136 (0.7)	124 (0.7)	1	1

Fig. 1 **A** Incidence and risk of newly developed diabetes in DAA- and interferon-treated patients. *Adjusted for age, sex, body mass index, hypertension, dyslipidemia, HCV genotype, HCV viral loads, aspartate aminotransferase, alanine transaminase, platelet counts, and FIB-4. *IFN* interferon-based patients, *DAA* direct-acting antivirals, *HR* hazard ratio. **B** Incidence and risk of newly developed cardio-cerebrovascular disease between DAA- and interferon-treated patients. **Adjusted for age, sex, body mass index, diabetes, hypertension, dyslipidemia, HCV genotype, HCV viral loads, aspartate aminotransferase, alanine transaminase, platelet counts, and FIB-4. *IFN* interferon-based patients, *DAA* direct-acting antivirals, *HR* hazard ratio

Table 4 Annual change of liver fibrosis measured by elastography and seromarkers in DAA-treated cohort

	Baseline	Year 1	Year 2	Year 3	Year 4	Year 5	P_{trend}
Liver stiffness, kPa	6.04 ± 4.81	6.24 ± 3.96	5.97 ± 3.48	5.98 ± 3.47	5.67 ± 2.93	5.51 ± 2.50	0.08
FIB-4	1.86 ± 0.92	1.86 ± 0.98	1.80 ± 0.81	1.79 ± 0.93	1.80 ± 0.84	1.76 ± 0.73	0.25
LOXL2, ng/ml	0.73 ± 2.12	1.29 ± 3.63	0.90 ± 2.71	0.60 ± 1.96	0.58 ± 3.10	0.35 ± 1.06	0.02
M2BPGi, COI	0.77 ± 0.47	0.76 ± 0.45	0.75 ± 0.42	0.75 ± 0.48	0.66 ± 0.46	0.71 ± 0.51	0.05

DAA directly acting antivirals, *FIB-4* fibrosis-4 index, *LOXL2* lysyl oxidase-like-2, *M2BPGi* Mac-2 binding protein glycosylation isomer

Table 5 Long-term change of quality of life in DAA-treated cohort

		Baseline	Year 1	Year 2	Year 3	Year 4	Year 5	P_{trend}
PCS	Physical health	78.0 ± 15.8	76.8 ± 15.3	76.5 ± 14.9	76.4 ± 15.7	75.7 ± 16.8	75.9 ± 16.5	0.17
PF	Physical functioning	87.9 ± 14.3	86.9 ± 14.5	86.6 ± 14.2	87.1 ± 16.4	85.3 ± 17.6	86.4 ± 16.4	0.23
RP	Role limitations because of physical health	82.6 ± 20.2	81.3 ± 19.7	79.1 ± 20.4	79.7 ± 20.1	78.8 ± 21.2	77.8 ± 21.2	0.02
BP	Bodily pain	84.4 ± 19.3	84.0 ± 18.0	83.4 ± 17.8	83.4 ± 19.5	82.3 ± 18.4	81.8 ± 18.8	0.14
GH	General health	65.3 ± 19.4	64.0 ± 18.9	62.7 ± 18.7	61.9 ± 19.2	63.0 ± 19.7	63.9 ± 19.8	0.33
MCS	Mental health	72.6 ± 16.9	71.3 ± 16.3	70.2 ± 16.4	69.4 ± 18.9	69.2 ± 17.6	69.0 ± 18.2	0.02
RE	Role limitations because of emotional problem	83.8 ± 20.7	83.4 ± 18.9	80.7 ± 19.5	80.6 ± 20.3	80.2 ± 21.8	78.3 ± 21.9	0.01
VT	Energy/vitality	67.8 ± 18.2	65.5 ± 18.6	66.6 ± 17.5	66.0 ± 18.7	65.8 ± 18.7	64.9 ± 20.0	0.22
MH	Mental health	74.2 ± 16.5	72.9 ± 16.5	71.1 ± 17.0	71.6 ± 17.6	71.4 ± 17.6	72.6 ± 17.1	0.21
SF	Social functioning	88.3 ± 15.3	86.4 ± 15.8	86.3 ± 15.5	86.0 ± 16.2	85.0 ± 17.0	85.2 ± 16.9	0.05

DAA directly acting antivirals

Regarding quality of life, a significant decrease in the SF-36 score was observed for role limitations due to physical health and emotional problems, whereas the other parameters related to physical and mental health were consistently maintained throughout the 5 years of follow-up (Table 5).

DISCUSSION

In the present study, we prospectively followed a DAA-treated cohort and compared it with a historically interferon-based control. We demonstrated that there was an equal risk of HCC and liver-related outcomes between the two groups after HCV eradication. A favorable

extrahepatic outcome, including the occurrence of diabetes and cardio-cerebrovascular disease, was noted, and liver fibrosis regression and quality of life maintenance continued after HCV was cured by DAAs.

HCV eradication via antiviral therapy greatly reduces the risk of HCC and liver-related events [6–8, 20]. In the early era of DAAs, a misunderstanding about increased post-SVR HCC risk existed, in part because of differences in patient characteristics and study designs. Compared with patients treated in the interferon era, DAA-treated patients are generally older, have more advanced liver fibrosis and comorbidities, and have shorter post-SVR follow-up periods. Additional studies have demonstrated the existence of equal benefits in reducing the risk of HCC in interferon- and DAA-treated patients after adjusting for potential confounders [10, 11]. Notably, most studies were retrospective, and a prospective study to address this issue is elusive. The present longitudinal follow-up study was in line with previous observations and confirmed that the risks of HCC and liver-related outcomes were similar between DAA- and interferon-treated patients after HCV cure.

Advanced liver fibrosis is the critical determinant of HCC in this HCV cohort as with other etiologies of liver disease [21, 22]. Liver biopsy is the gold standard for evaluating liver fibrosis. However, it is limited by the invasiveness, sampling and interpretation variability, and the unavailability of repeat judgments. Liver stiffness measurement by imaging modalities and serum-based fibrosis markers has been reasonably and widely adopted to replace histology in the clinical setting. Nevertheless, noninvasive scores and liver stiffness measurements by transient elastography methods are not accurate in the diagnosis of fibrosis regression after HCV eradication, and the cutoff values of different fibrotic stages in the post-SVR status remain unclear [23]. The development of new noninvasive serum markers to represent liver fibrosis is important. As collagen type I increases during hepatic fibrogenesis, LOXL2 may modulate type 1 collagen, in turn leading to an elevation in its serum level in patients with liver fibrosis [24]. On the other hand, fibrosis leads to specific modifications of the glycosylation and sugar chain structure of M2BP. As a result, a high M2BPGi level serves as an indicator of liver disease severity and a predictor of liver-related outcomes. Assuming that histological fibrosis continues to resolve in the current DAA cohort, we observed that the FIB-4 value did not decrease consistently and may not be suitable for serving as a surrogate for liver fibrosis in the post-SVR cohort. Moreover, the serum level of LOXL2 may outperform M2BP, which was consistently reduced at up to 5 years of follow-up. M2BPGi levels are a predictor for HCC with different etiologies [25–27]. Further studies are needed to explore the association of serum LOXL2 levels with HCC risk.

Post-SVR surveillance of HCC in terms of the target population and screening strategies is a critical issue. All international societies agree to follow cirrhosis patients indefinitely, but discrepancies exist in noncirrhotic patients across regional guidelines [10]. The three HCC patients in the DAA cohort were noncirrhotic at baseline but had progressively elevated liver stiffness. These findings echo the recent statement of the European Association for the Study of the Liver (EASL) that subjects with borderline advanced liver fibrosis should be considered for the existence of metabolic dysfunction-associated steatotic liver disease or alcoholic liver disease, and liver fibrosis should be followed by noninvasive testing (NIT) after achieving SVR [28]. Notably, even if clinicians have identified high-risk targets to be followed, host genetic predispositions and alternations, healthcare system provider barriers, and patient adherence are some of the the hurdles encountered and remain unmet needs for post-CVR HCC surveillance [10, 29].

HCV eradication improves glycemic control [30] and reduces diabetes-related complications [31]. During the prospective follow-up period, we noticed a minimal incidence of new diabetes and cardio-cerebrovascular disease. The finding regarding a more favorable extrahepatic outcome in the DAA cohort should be considered with caveats. Even though we adjusted age, sex, BMI, hypertension, DM, and dyslipidemia while exploring the cardio-cerebrovascular disease, selection bias between groups may still exist. On the other hand, neuropsychiatric disturbance is one of the extrahepatic presentations of CHC. HCV eradication improves quality of

life [19]. Nevertheless, all the patient-reported outcomes were restricted to short-term reports after HCV eradication [9], and long-term observations of QoL are lacking. While the subjects were fighting against the mental and physical aging process, we observed that most items on the SF-36 were maintained up to 5 years after HCV cure. This is by far the longest follow-up study to evaluate the long-term presentation of QoL after HCV eradication.

The current study was limited by the small sample size. It is impracticable to enroll prospective interferon-treated controls, and the historical control may have unmeasured confounders. Unadjusted confounders such as smoking, lipid profiles, hypertension management, and glycemic control may constrain the comparison of the cardio-cerebrovascular risk between groups. The registry study was designed to strictly and prospectively follow DAA-treated patients after they completed phase III studies in Taiwan and Korea. The results reflected the incidences of hepatic and extrahepatic events after HCV cure. In conclusion, HCV eradication by DAAs improved liver- and non-liver-related outcomes, promoted liver fibrosis regression, and maintained quality of life after HCV cure.

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Declarations

Conflict of Interests. Ming-Lung Yu: Research support from AbbVie, Abbott, BMS, Gilead, Merck and Roche. Consultant for AbbVie, Abbott, Ascletis, BMS, Gilead Sciences, J&J, Merck, Novartis, Pharmaessential and Roche. Speaker for AbbVie, Abbott, Ascletis, BMS, Gilead Sciences, Merck, Pharmaessential and Roche. Chung-Feng Huang: Speaker for AbbVie, BMS, Gilead Sciences, Merck, and Roche. Young-Suk Lim: Research support and consultant for Gilead Sciences. Young-Seok Kim: Speaker, consultant and grant support for Gilead Sciences. Jeong Huo, Rong-Nan Chien, Yang-Hyun Baek, Jia-Horng Kao, Ju-Hyun Kim, Ting-Tsang Chang, Kwan-Soo Byun, Jyh-Jou Chen, Sook-Hyang Jeong, Tsung-Hui Hu, Cheng-Yuan Peng, Won-Young Tak, Hrong-Yuan Wang, Seung-Kew Yoon, I-Shyan Sheen, Youn-Jae Lee, Yu-Chun Hsu, Pei-Chien Tsai, Ming-Lun Yeh, Sang-Hoon Ahn, Chai-Yen Dai, Seung-Woon Paik, Jee-Fu Huang, Yoon-Jun Kim, and Wan-Long Chuang have nothing to disclose.

Ethical Approval. The study was approved by the institutional review board of Kaohsiung Medical University Hospital (KMUHIRB-F(I)-20160082), which conforms to the guidelines of the International Conference on Harmonization for Good Clinical Practice and was performed in accordance with the Helsinki Declaration of 1964 and its later amendments. All patients provided written informed consent.

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